

SEQUENCE LISTING

<110> KAO CORPORATION

<120> Mutant Bacillus

<130> KS0816

<150> JP 2004-062852

<151> 2004.03.05

<160> 28

<170> PatentIn Ver. 3.1

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<212> PRT

<213> Bacillus subtilis

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Arg Phe Gly Leu Asp Asp Gly Arg Thr Arg Thr Leu Glu Glu Val Gly
325 330 335

Lys Val Phe Gly Val Thr Arg Glu Arg Ile Arg Gln Ile Glu Ala Lys
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 35 40 45

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 65 70 75 80

Ile Arg Leu Ala Met Tyr Val Gly Glu Asn Gly Tyr Ala Thr Asn Pro
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 100 105 110

Asn Asp Met Tyr Val Ile Val Asp Trp His Val His Ala Pro Gly Asp
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 130 135 140

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Pro Asn Trp Ser Gln Arg Pro Asp Leu Ala Ala Asp Asn Pro Ile Asp
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Pro Gly Pro Asp His Val Trp Ala Pro Glu Glu Leu Ser Leu Ser Gly
340 345 350

Glu Tyr Val Arg Ala Arg Ile Lys Gly Val Asn Tyr Glu Pro Ile Asp
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Arg Thr Lys Tyr Thr Lys Val Leu Trp Asp Phe Asn Asp Gly Thr Lys
370 375 380

Gln Gly Phe Gly Val Asn Ser Asp Ser Pro Asn Lys Glu Leu Ile Ala
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Val Asp Asn Glu Asn Asn Thr Leu Lys Val Ser Gly Leu Asp Val Ser
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Asn Asp Val Ser Asp Gly Asn Phe Trp Ala Asn Ala Arg Leu Ser Ala
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Asn Gly Trp Gly Lys Ser Val Asp Ile Leu Gly Ala Glu Lys Leu Thr
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Met Asp Val Ile Val Asp Glu Pro Thr Thr Val Ala Ile Ala Ala Ile
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Phe His Glu Glu Asp Asn Asn Met Asn Asn Ile Ile Leu Phe Val Gly
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Thr Asp Ala Ala Asp Val Ile Tyr Leu Asp Asn Ile Lys Val Ile Gly
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565 570 575

Gly Glu Ser Gly Val Lys Thr Ala Leu Thr Ile Glu Glu Ala Asn Gly
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Ser Asn Ala Leu Ser Trp Glu Phe Gly Tyr Pro Glu Val Lys Pro Ser
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Pro Thr Asn Gly Tyr Trp Val Gln Ala Pro Lys Thr Tyr Thr Ile Asn
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Phe Asp Glu Leu Glu Glu Ala Asn Gln Val Asn Gly Leu Tyr His Tyr
675 680 685

Glu Val Lys Ile Asn Val Arg Asp Ile Thr Asn Ile Gln Asp Asp Thr
690 695 700

Leu Leu Arg Asn Met Met Ile Ile Phe Ala Asp Val Glu Ser Asp Phe

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 Phe Glu Gly Ala Ala Thr Thr Glu Pro Val Glu Pro Glu Pro Val Asp
 735 740 745

cct ggc gaa gag acg cca cct gtc gat gag aag gaa gcg aaa aaa gaa 2945
 Pro Gly Glu Glu Thr Pro Pro Val Asp Glu Lys Glu Ala Lys Lys Glu
 750 755 760

caa aaa gaa gca gag aaa gaa gag aaa gaa gca gta aaa gaa gaa aag 2993
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 765 770 775

aaa gaa gct aaa gaa gaa aag aaa gca gtc aaa aat gag gct aag aaa 3041
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 780 785 790

aaa taatctatta aactagttat agggttatct aaaggtctga ttagatctt 3094
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Pro Asn Trp Ser Gln Arg Pro Asp Leu Ala Ala Asp Asn Pro Ile Asp
210 215 220

Asp His His Thr Met Tyr Thr Val His Phe Tyr Thr Gly Ser His Ala
225 230 235 240

Ala Ser Thr Glu Ser Tyr Pro Pro Glu Thr Pro Asn Ser Glu Arg Gly
245 250 255

Asn Val Met Ser Asn Thr Arg Tyr Ala Leu Glu Asn Gly Val Ala Val
260 265 270

Phe Ala Thr Glu Trp Gly Thr Ser Gln Ala Asn Gly Asp Gly Gly Pro
275 280 285

Tyr Phe Asp Glu Ala Asp Val Trp Ile Glu Phe Leu Asn Glu Asn Asn
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340 345 350

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355 360 365

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370 375 380

Gln Gly Phe Gly Val Asn Gly Asp Ser Pro Val Glu Asp Val Val Ile
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405 410 415

Asp Val Ser Glu Gly Asn Tyr Trp Ala Asn Ala Arg Leu Ser Ala Asp
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Gly Trp Gly Lys Ser Val Asp Ile Leu Gly Ala Glu Lys Leu Thr Met
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Gln Gly Pro Ser Ala Asn Trp Val Asn Pro Asn Arg Ala Ile Lys Val
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485 490 495

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Ser Gly Val Lys Thr Ala Leu Thr Ile Glu Glu Ala Asn Gly Ser Asn
580 585 590

Ala Leu Ser Trp Glu Phe Gly Tyr Pro Glu Val Lys Pro Ser Asp Asn
595 600 605

Trp Ala Thr Ala Pro Arg Leu Asp Phe Trp Lys Ser Asp Leu Val Arg
610 615 620

Gly Glu Asn Asp Tyr Val Thr Phe Asp Phe Tyr Leu Asp Pro Val Arg

625 630 635 640

Ala Thr Glu Gly Ala Met Asn Ile Asn Leu Val Phe Gln Pro Pro Thr
 645 650 655

Asn Gly Tyr Trp Val Gln Ala Pro Lys Thr Tyr Thr Ile Asn Phe Asp
 660 665 670

Glu Leu Glu Glu Ala Asn Gln Val Asn Gly Leu Tyr His Tyr Glu Val
 675 680 685

Lys Ile Asn Val Arg Asp Ile Thr Asn Ile Gln Asp Asp Thr Leu Leu
 690 695 700

Arg Asn Met Met Ile Ile Phe Ala Asp Val Glu Ser Asp Phe Ala Gly
 705 710 715 720

Arg Val Phe Val Asp Asn Val Arg Phe Glu Gly Ala Ala Thr Thr Glu
 725 730 735

Pro Val Glu Pro Glu Pro Val Asp Pro Gly Glu Glu Thr Pro Pro Val
 740 745 750

Asp Glu Lys Glu Ala Lys Lys Glu Gln Lys Glu Ala Glu Lys Glu Glu
 755 760 765

Lys Glu Ala Val Lys Glu Glu Lys Lys Glu Ala Lys Glu Glu Lys Lys
 770 775 780

Ala Ile Lys Asn Glu Ala Thr Lys Lys
 785 790

<210> 7
 <211> 3332
 <212> DNA
 <213> Bacillus sp. KSM-64

<220>

<221> CDS
 <222> (610).. (3075)
 <223>

<220>
 <221> sig_peptide
 <222> (610).. (696)
 <223>

<220>
 <221> mat_peptide
 <222> (697).. (3075)
 <223>

<400> 7
 agtacttacc atttttagagt caaaagatag aagccaagca ggatttgccg atgcaaccgg 60
 cttatattta gaggggaattt ctttttaaatt tgaatacggg ataaaatcag gtaaacaggt 120
 cctgatttta tttttttgaa tttttttgag aactaaagat tgaaatagaa gtagaagaca 180
 acggacataa gaaaattgta ttagttttta ttatagaaaa cgcttttcta taattattta 240
 tacctagaac gaaaatactg tttcgaaagc ggtttactat aaaaccttat attccggctc 300
 tttttttaaa caggggggtga aaattcactc tagtattcta atttcaacat gctataataa 360
 atttgaaga cgcaatatac atcttttttt tatgatattt gtaagcgggt aaccttgtgc 420
 tatatgccga tttaggaagg gggtagattg agtcaagtag tcataattta gataacttat 480
 aagttgttga gaagcaggag agaatctggg ttactcacia gttttttaaa acattatcga 540
 aagcactttc ggttatgctt atgaatttag ctatttgatt caattacttt aataatttta 600
 ggaggtaat atg atg tta aga aag aaa aca aag cag ttg att tct tcc att 651
 Met Met Leu Arg Lys Lys Thr Lys Gln Leu Ile Ser Ser Ile
 -25 -20
 ctt att tta gtt tta ctt cta tct tta ttt ccg aca gct ctt gca gca 699
 Leu Ile Leu Val Leu Leu Leu Ser Leu Phe Pro Thr Ala Leu Ala Ala
 -15 -10 -5 -1 1
 gaa gga aac act cgt gaa gac aat ttt aaa cat tta tta ggt aat gac 747
 Glu Gly Asn Thr Arg Glu Asp Asn Phe Lys His Leu Leu Gly Asn Asp
 5 10 15
 aat gtt aaa cgc cct tct gag gct ggc gca tta caa tta caa gaa gtc 795
 Asn Val Lys Arg Pro Ser Glu Ala Gly Ala Leu Gln Leu Gln Glu Val

20	25	30	
gat gga caa atg aca tta gta gat caa cat gga gaa aaa att caa tta			843
Asp Gly Gln Met Thr Leu Val Asp Gln His Gly Glu Lys Ile Gln Leu			
35	40	45	
cgt gga atg agt aca cac gga tta caa tgg ttt cct gag atc ttg aat			891
Arg Gly Met Ser Thr His Gly Leu Gln Trp Phe Pro Glu Ile Leu Asn			
50	55	60	65
gat aac gca tac aaa gct ctt gct aac gat tgg gaa tca aat atg att			939
Asp Asn Ala Tyr Lys Ala Leu Ala Asn Asp Trp Glu Ser Asn Met Ile			
70	75	80	
cgt cta gct atg tat gtc ggt gaa aat ggc tat gct tca aat cca gag			987
Arg Leu Ala Met Tyr Val Gly Glu Asn Gly Tyr Ala Ser Asn Pro Glu			
85	90	95	
tta att aaa agc aga gtc att aaa gga ata gat ctt gct att gaa aat			1035
Leu Ile Lys Ser Arg Val Ile Lys Gly Ile Asp Leu Ala Ile Glu Asn			
100	105	110	
gac atg tat gtc atc gtt gat tgg cat gta cat gca cct ggt gat cct			1083
Asp Met Tyr Val Ile Val Asp Trp His Val His Ala Pro Gly Asp Pro			
115	120	125	
aga gat ccc gtt tac gct gga gca gaa gat ttc ttt aga gat att gca			1131
Arg Asp Pro Val Tyr Ala Gly Ala Glu Asp Phe Phe Arg Asp Ile Ala			
130	135	140	145
gca tta tat cct aac aat cca cac att att tat gag tta gcg aat gag			1179
Ala Leu Tyr Pro Asn Asn Pro His Ile Ile Tyr Glu Leu Ala Asn Glu			
150	155	160	
cca agt agt aac aat aat ggt gga gct ggg att cca aat aat gaa gaa			1227
Pro Ser Ser Asn Asn Asn Gly Gly Ala Gly Ile Pro Asn Asn Glu Glu			
165	170	175	
ggt tgg aat gcg gta aaa gaa tac gct gat cca att gta gaa atg tta			1275
Gly Trp Asn Ala Val Lys Glu Tyr Ala Asp Pro Ile Val Glu Met Leu			
180	185	190	
cgt gat agc ggg aac gca gat gac aat att atc att gtg ggt agt cca			1323
Arg Asp Ser Gly Asn Ala Asp Asp Asn Ile Ile Ile Val Gly Ser Pro			
195	200	205	
aac tgg agt cag cgt cct gac tta gca gct gat aat cca att gat gat			1371
Asn Trp Ser Gln Arg Pro Asp Leu Ala Ala Asp Asn Pro Ile Asp Asp			
210	215	220	225

cac cat aca atg tat act gtt cac ttc tac act ggt tca cat gct gct His His Thr Met Tyr Thr Val His Phe Tyr Thr Gly Ser His Ala Ala 230 235 240	1419
tca act gaa agc tat ccg cct gaa act cct aac tct gaa aga gga aac Ser Thr Glu Ser Tyr Pro Pro Glu Thr Pro Asn Ser Glu Arg Gly Asn 245 250 255	1467
gta atg agt aac act cgt tat gcg tta gaa aac gga gta gca gta ttt Val Met Ser Asn Thr Arg Tyr Ala Leu Glu Asn Gly Val Ala Val Phe 260 265 270	1515
gca aca gag tgg gga act agc caa gca aat gga gat ggt ggt cct tac Ala Thr Glu Trp Gly Thr Ser Gln Ala Asn Gly Asp Gly Gly Pro Tyr 275 280 285	1563
ttt gat gaa gca gat gta tgg att gag ttt tta aat gaa aac aac att Phe Asp Glu Ala Asp Val Trp Ile Glu Phe Leu Asn Glu Asn Asn Ile 290 295 300 305	1611
agc tgg gct aac tgg tct tta acg aat aaa aat gaa gta tct ggt gca Ser Trp Ala Asn Trp Ser Leu Thr Asn Lys Asn Glu Val Ser Gly Ala 310 315 320	1659
ttt aca cca ttc gag tta ggt aag tct aac gca aca agt ctt gac cca Phe Thr Pro Phe Glu Leu Gly Lys Ser Asn Ala Thr Ser Leu Asp Pro 325 330 335	1707
ggg cca gac caa gta tgg gta cca gaa gag tta agt ctt tct gga gaa Gly Pro Asp Gln Val Trp Val Pro Glu Glu Leu Ser Leu Ser Gly Glu 340 345 350	1755
tat gta cgt gct cgt att aaa ggt gtg aac tat gag cca atc gac cgt Tyr Val Arg Ala Arg Ile Lys Gly Val Asn Tyr Glu Pro Ile Asp Arg 355 360 365	1803
aca aaa tac acg aaa gta ctt tgg gac ttt aat gat gga acg aag caa Thr Lys Tyr Thr Lys Val Leu Trp Asp Phe Asn Asp Gly Thr Lys Gln 370 375 380 385	1851
gga ttt gga gtg aat gga gat tct cca gtt gaa gat gta gtt att gag Gly Phe Gly Val Asn Gly Asp Ser Pro Val Glu Asp Val Val Ile Glu 390 395 400	1899
aat gaa gcg ggc gct tta aaa ctt tca gga tta gat gca agt aat gat Asn Glu Ala Gly Ala Leu Lys Leu Ser Gly Leu Asp Ala Ser Asn Asp 405 410 415	1947

gtt tct gaa ggt aat tac tgg gct aat gct cgt ctt tct gcc gac ggt Val Ser Glu Gly Asn Tyr Trp Ala Asn Ala Arg Leu Ser Ala Asp Gly 420 425 430	1995
tgg gga aaa agt gtt gat att tta ggt gct gaa aaa ctt act atg gat Trp Gly Lys Ser Val Asp Ile Leu Gly Ala Glu Lys Leu Thr Met Asp 435 440 445	2043
gtg att gtt gat gag ccg acc acg gta tca att gct gca att cca caa Val Ile Val Asp Glu Pro Thr Thr Val Ser Ile Ala Ala Ile Pro Gln 450 455 460 465	2091
ggg cca tca gcc aat tgg gtt aat cca aat cgt gca att aag gtt gag Gly Pro Ser Ala Asn Trp Val Asn Pro Asn Arg Ala Ile Lys Val Glu 470 475 480	2139
cca act aat ttc gta ccg tta gga gat aag ttt aaa gcg gaa tta act Pro Thr Asn Phe Val Pro Leu Gly Asp Lys Phe Lys Ala Glu Leu Thr 485 490 495	2187
ata act tca gct gac tct cca tcg tta gaa gct att gcg atg cat gct Ile Thr Ser Ala Asp Ser Pro Ser Leu Glu Ala Ile Ala Met His Ala 500 505 510	2235
gaa aat aac aac atc aac aac atc att ctt ttt gta gga act gaa ggt Glu Asn Asn Asn Ile Asn Asn Ile Ile Leu Phe Val Gly Thr Glu Gly 515 520 525	2283
gct gat gtt atc tat tta gat aac att aaa gta att gga aca gaa gtt Ala Asp Val Ile Tyr Leu Asp Asn Ile Lys Val Ile Gly Thr Glu Val 530 535 540 545	2331
gaa att cca gtt gtt cat gat cca aaa gga gaa gct gtt ctt cct tct Glu Ile Pro Val Val His Asp Pro Lys Gly Glu Ala Val Leu Pro Ser 550 555 560	2379
gtt ttt gaa gac ggt aca cgt caa ggt tgg gac tgg gct gga gag tct Val Phe Glu Asp Gly Thr Arg Gln Gly Trp Asp Trp Ala Gly Glu Ser 565 570 575	2427
ggt gtg aaa aca gct tta aca att gaa gaa gca aac ggt tct aac gcg Gly Val Lys Thr Ala Leu Thr Ile Glu Glu Ala Asn Gly Ser Asn Ala 580 585 590	2475
tta tca tgg gaa ttt gga tac cca gaa gta aaa cct agt gat aac tgg Leu Ser Trp Glu Phe Gly Tyr Pro Glu Val Lys Pro Ser Asp Asn Trp 595 600 605	2523
gca aca gct cca cgt tta gat ttc tgg aaa tct gac ttg gtt cgc ggt	2571

Ala Thr Ala Pro Arg Leu Asp Phe Trp Lys Ser Asp Leu Val Arg Gly	
610 615 620 625	
gaa aat gat tat gta act ttt gat ttc tat cta gat cca gtt cgt gca	2619
Glu Asn Asp Tyr Val Thr Phe Asp Phe Tyr Leu Asp Pro Val Arg Ala	
630 635 640	
aca gaa ggc gca atg aat atc aat tta gta ttc cag cca cct act aac	2667
Thr Glu Gly Ala Met Asn Ile Asn Leu Val Phe Gln Pro Pro Thr Asn	
645 650 655	
ggg tat tgg gta caa gca cca aaa acg tat acg att aac ttt gat gaa	2715
Gly Tyr Trp Val Gln Ala Pro Lys Thr Tyr Thr Ile Asn Phe Asp Glu	
660 665 670	
tta gag gaa gcg aat caa gta aat ggt tta tat cac tat gaa gtg aaa	2763
Leu Glu Glu Ala Asn Gln Val Asn Gly Leu Tyr His Tyr Glu Val Lys	
675 680 685	
att aac gta aga gat att aca aac att caa gat gac acg tta cta cgt	2811
Ile Asn Val Arg Asp Ile Thr Asn Ile Gln Asp Asp Thr Leu Leu Arg	
690 695 700 705	
aac atg atg atc att ttt gca gat gta gaa agt gac ttt gca ggg aga	2859
Asn Met Met Ile Ile Phe Ala Asp Val Glu Ser Asp Phe Ala Gly Arg	
710 715 720	
gtc ttt gta gat aat gtt cgt ttt gag ggg gct gct act act gag ccg	2907
Val Phe Val Asp Asn Val Arg Phe Glu Gly Ala Ala Thr Thr Glu Pro	
725 730 735	
gtt gaa cca gag cca gtt gat cct ggc gaa gag acg ccg cct gtc gat	2955
Val Glu Pro Glu Pro Val Asp Pro Gly Glu Glu Thr Pro Pro Val Asp	
740 745 750	
gag aag gaa gcg aaa aaa gaa caa aaa gaa gca gag aaa gaa gag aaa	3003
Glu Lys Glu Ala Lys Lys Glu Gln Lys Glu Ala Glu Lys Glu Glu Lys	
755 760 765	
gaa gca gta aaa gaa gaa aag aaa gaa gct aaa gaa gaa aag aaa gca	3051
Glu Ala Val Lys Glu Glu Lys Lys Glu Ala Lys Glu Glu Lys Lys Ala	
770 775 780 785	
atc aaa aat gag gct acg aaa aaa taatctaata aactagttat agggttatct	3105
Ile Lys Asn Glu Ala Thr Lys Lys	
790	
aaaggctctga tgcagatctt ttagataaacc tttttttgca taactggaca tagaatggtt	3165

attaaagaaa gcaagggtgtt tatacgatat taaaaaggta gcgattttta attgaaacct 3225

ttaataatgt ctgtgatag aatgatgaag taatttaaga gggggaaacg aagtgaaaac 3285

ggaaatttct agtagaagaa aaacagacca agaaatactg caagctt 3332

<210> 8

<211> 20

<212> DNA

<213> artificial sequence

<220>

<223> Oligonucleotide as PCR primer designed from nucleotide sequence of sigA in *Bacillus subtilis*

<400> 8

atggctgata aacaaacca 20

<210> 9

<211> 20

<212> DNA

<213> artificial sequence

<220>

<223> Oligonucleotide as PCR primer designed from nucleotide sequence of sigA in *Bacillus subtilis*

<400> 9

caccacaatg ttcatttgca 20

<210> 10

<211> 21

<212> DNA

<213> artificial sequence

<220>

<223> Oligonucleotide as PCR primer designed from nucleotide sequence of the upstream region of sigH in *Bacillus subtilis*

<400> 10

acagcctttc ttcctcattc t 21

<210> 11

<211> 42

<212> DNA

<213> artificial sequence

<220>

<223> Oligonucleotide as PCR primer; its 3'-portion designed from nucleotide sequence of the upstream region of sigH in *Bacillus subtilis* and its 5'-portion designed from nucleotide sequence of sigA in *Bacillus subtilis*

<400> 11

cgtgggtttg tttatcagcc attccgatcc ccccggcgca cg 42

<210> 12

<211> 21

<212> DNA

<213> artificial sequence

<220>

<223> Oligonucleotide as PCR primer designed from nucleotide sequence of the upstream region of sigF in *Bacillus subtilis*

<400> 12

gctgatagaa cgtgacacgg g 21

<210> 13

<211> 42

<212> DNA

<213> artificial sequence

<220>

<223> Oligonucleotide as PCR primer; its 3'-portion designed from nucleotide sequence of the upstream region of sigF in *Bacillus subtilis* and its 5'-portion designed from nucleotide sequence of sigA in *Bacillus subtilis*

<400> 13

cgtgggtttg tttatcagcc atgctcattc ctccttgata tg 42

<210> 14

<211> 19

<212> DNA

<213> artificial sequence

<220>

<223> Oligonucleotide as PCR primer designed from nucleotide sequence of plasmid pC194

<400> 14
caactaaagc acccattag 19

<210> 15
<211> 44
<212> DNA
<213> artificial sequence

<220>

<223> Oligonucleotide as PCR primer; its 3'-portion designed from nucleotide sequence of plasmid pC194 and its 5'-portion designed from nucleotide sequence of sigA in *Bacillus subtilis*

<400> 15
catttgcaaa tgaacattgt ggtgcttctt caactaacgg ggca 44

<210> 16
<211> 20
<212> DNA
<213> artificial sequence

<220>

<223> Oligonucleotide as PCR primer designed from nucleotide sequence of sigA in *Bacillus subtilis*; the sequence containing a nucleotide substitution for destroying the initiation codon of sigA

<400> 16
atagctgata aacaaaccca 20

<210> 17
<211> 42
<212> DNA
<213> artificial sequence

<220>

<223> Oligonucleotide as PCR primer; its 3'-portion designed from nucleotide sequence of the upstream region of sigF in *Bacillus subtilis* and its 5'-portion designed from nucleotide sequence of sigA in *Bacillus subtilis*; the sequence containing a nucleotide substitution for destroying the initiation codon of sigA

<400> 17
cgtgggtttg tttatcagct atgctcattc ctccttgata tg 42

<210> 18

<211> 1795
 <212> DNA
 <213> Bacillus sp. KSM-K38

<220>
 <221> CDS
 <222> (212).. (1714)
 <223>

<220>
 <221> sig_peptide
 <222> (212).. (274)
 <223>

<220>
 <221> mat_peptide
 <222> (275).. (1714)
 <223>

<400> 18

caggccagcc aaagtagcca ccaactaagt aacatcgatt caggataaaa gtatgcgaaa	60
cgatgcgcaa aactgcgcaa ctactagcac tcttcaggga ctaaaccacc ttttttccaa	120
aaatgacatc atataaaciaa atttgtctac caatcactat ttaaagctgt ttaatgatata	180
tgtaagcggt atcattaaaa ggaggtatgt g atg aga aga tgg gta gta gca	232
Met Arg Arg Trp Val Val Ala	
-20 -15	
atg ttg gca gtg tta ttt tta ttt cct tcg gta gta gtt gca gat gga	280
Met Leu Ala Val Leu Phe Leu Phe Pro Ser Val Val Val Ala Asp Gly	
-10 -5 -1 1	
ttg aac ggt acg atg atg cag tat tat gag tgg cat ttg gaa aac gac	328
Leu Asn Gly Thr Met Met Gln Tyr Tyr Glu Trp His Leu Glu Asn Asp	
5 10 15	
ggg cag cat tgg aat cgg ttg cac gat gat gcc gca gct ttg agt gat	376
Gly Gln His Trp Asn Arg Leu His Asp Asp Ala Ala Ala Leu Ser Asp	
20 25 30	
gct ggt att aca gct att tgg att ccg cca gcc tac aaa ggt aat agt	424
Ala Gly Ile Thr Ala Ile Trp Ile Pro Pro Ala Tyr Lys Gly Asn Ser	
35 40 45 50	
cag gcg gat gtt ggg tac ggt gca tac gat ctt tat gat tta gga gag	472
Gln Ala Asp Val Gly Tyr Gly Ala Tyr Asp Leu Tyr Asp Leu Gly Glu	

55	60	65	
ttc aat caa aag ggt act gtt cga acg aaa tac gga act aag gca cag			520
Phe Asn Gln Lys Gly Thr Val Arg Thr Lys Tyr Gly Thr Lys Ala Gln			
70	75	80	
ctt gaa cga gct att ggg tcc ctt aaa tct aat gat atc aat gta tac			568
Leu Glu Arg Ala Ile Gly Ser Leu Lys Ser Asn Asp Ile Asn Val Tyr			
85	90	95	
gga gat gtc gtg atg aat cat aaa atg gga gct gat ttt acg gag gca			616
Gly Asp Val Val Met Asn His Lys Met Gly Ala Asp Phe Thr Glu Ala			
100	105	110	
gtg caa gct gtt caa gta aat cca acg aat cgt tgg cag gat att tca			664
Val Gln Ala Val Gln Val Asn Pro Thr Asn Arg Trp Gln Asp Ile Ser			
115	120	125	130
ggt gcc tac acg att gat gcg tgg acg ggt ttc gac ttt tca ggg cgt			712
Gly Ala Tyr Thr Ile Asp Ala Trp Thr Gly Phe Asp Phe Ser Gly Arg			
135	140	145	
aac aac gcc tat tca gat ttt aag tgg aga tgg ttc cat ttt aat ggt			760
Asn Asn Ala Tyr Ser Asp Phe Lys Trp Arg Trp Phe His Phe Asn Gly			
150	155	160	
gtt gac tgg gat cag cgc tat caa gaa aat cat att ttc cgc ttt gca			808
Val Asp Trp Asp Gln Arg Tyr Gln Glu Asn His Ile Phe Arg Phe Ala			
165	170	175	
aat acg aac tgg aac tgg cga gtg gat gaa gag aac ggt aat tat gat			856
Asn Thr Asn Trp Asn Trp Arg Val Asp Glu Glu Asn Gly Asn Tyr Asp			
180	185	190	
tac ctg tta gga tcg aat atc gac ttt agt cat cca gaa gta caa gat			904
Tyr Leu Leu Gly Ser Asn Ile Asp Phe Ser His Pro Glu Val Gln Asp			
195	200	205	210
gag ttg aag gat tgg ggt agc tgg ttt acc gat gag tta gat ttg gat			952
Glu Leu Lys Asp Trp Gly Ser Trp Phe Thr Asp Glu Leu Asp Leu Asp			
215	220	225	
ggt tat cgt tta gat gct att aaa cat att cca ttc tgg tat aca tct			1000
Gly Tyr Arg Leu Asp Ala Ile Lys His Ile Pro Phe Trp Tyr Thr Ser			
230	235	240	
gat tgg gtt cgg cat cag cgc aac gaa gca gat caa gat tta ttt gtc			1048
Asp Trp Val Arg His Gln Arg Asn Glu Ala Asp Gln Asp Leu Phe Val			
245	250	255	

gta ggg gaa tat tgg aag gat gac gta ggt gct ctc gaa ttt tat tta Val Gly Glu Tyr Trp Lys Asp Asp Val Gly Ala Leu Glu Phe Tyr Leu 260 265 270	1096
gat gaa atg aat tgg gag atg tct cta ttc gat gtt cca ctt aat tat Asp Glu Met Asn Trp Glu Met Ser Leu Phe Asp Val Pro Leu Asn Tyr 275 280 285 290	1144
aat ttt tac cgg gct tca caa caa ggt gga agc tat gat atg cgt aat Asn Phe Tyr Arg Ala Ser Gln Gln Gly Gly Ser Tyr Asp Met Arg Asn 295 300 305	1192
att tta cga gga tct tta gta gaa gcg cat ccg atg cat gca gtt acg Ile Leu Arg Gly Ser Leu Val Glu Ala His Pro Met His Ala Val Thr 310 315 320	1240
ttt gtt gat aat cat gat act cag cca ggg gag tca tta gag tca tgg Phe Val Asp Asn His Asp Thr Gln Pro Gly Glu Ser Leu Glu Ser Trp 325 330 335	1288
gtt gct gat tgg ttt aag cca ctt gct tat gcg aca att ttg acg cgt Val Ala Asp Trp Phe Lys Pro Leu Ala Tyr Ala Thr Ile Leu Thr Arg 340 345 350	1336
gaa ggt ggt tat cca aat gta ttt tac ggt gat tac tat ggg att cct Glu Gly Gly Tyr Pro Asn Val Phe Tyr Gly Asp Tyr Tyr Gly Ile Pro 355 360 365 370	1384
aac gat aac att tca gct aaa aaa gat atg att gat gag ctg ctt gat Asn Asp Asn Ile Ser Ala Lys Lys Asp Met Ile Asp Glu Leu Leu Asp 375 380 385	1432
gca cgt caa aat tac gca tat ggc acg cag cat gac tat ttt gat cat Ala Arg Gln Asn Tyr Ala Tyr Gly Thr Gln His Asp Tyr Phe Asp His 390 395 400	1480
tgg gat gtt gta gga tgg act agg gaa gga tct tcc tcc aga cct aat Trp Asp Val Val Gly Trp Thr Arg Glu Gly Ser Ser Ser Arg Pro Asn 405 410 415	1528
tca ggc ctt gcg act att atg tog aat gga cct ggt ggt tcc aag tgg Ser Gly Leu Ala Thr Ile Met Ser Asn Gly Pro Gly Gly Ser Lys Trp 420 425 430	1576
atg tat gta gga cgt cag aat gca gga caa aca tgg aca gat tta act Met Tyr Val Gly Arg Gln Asn Ala Gly Gln Thr Trp Thr Asp Leu Thr 435 440 445 450	1624

ggt aat aac gga gcg tcc gtt aca att aat ggc gat gga tgg ggc gaa 1672
 Gly Asn Asn Gly Ala Ser Val Thr Ile Asn Gly Asp Gly Trp Gly Glu
 455 460 465

ttc ttt acg aat gga gga tct gta tcc gtg tac gtg aac caa taacaaaaa 1723
 Phe Phe Thr Asn Gly Gly Ser Val Ser Val Tyr Val Asn Gln
 470 475 480

gccttgagaa gggattcctc cctaactcaa ggctttcttt atgtcgctta gctttacgct 1783
 tctacgactt tg 1795

<210> 19
 <211> 480
 <212> PRT
 <213> Bacillus sp. KSM-K38

<400> 19

Asp Gly Leu Asn Gly Thr Met Met Gln Tyr Tyr Glu Trp His Leu Glu
 1 5 10 15

Asn Asp Gly Gln His Trp Asn Arg Leu His Asp Asp Ala Ala Ala Leu
 20 25 30

Ser Asp Ala Gly Ile Thr Ala Ile Trp Ile Pro Pro Ala Tyr Lys Gly
 35 40 45

Asn Ser Gln Ala Asp Val Gly Tyr Gly Ala Tyr Asp Leu Tyr Asp Leu
 50 55 60

Gly Glu Phe Asn Gln Lys Gly Thr Val Arg Thr Lys Tyr Gly Thr Lys
 65 70 75 80

Ala Gln Leu Glu Arg Ala Ile Gly Ser Leu Lys Ser Asn Asp Ile Asn
 85 90 95

Val Tyr Gly Asp Val Val Met Asn His Lys Met Gly Ala Asp Phe Thr
 100 105 110

Glu Ala Val Gln Ala Val Gln Val Asn Pro Thr Asn Arg Trp Gln Asp

115	120	125
Ile Ser Gly Ala Tyr Thr	Ile Asp Ala Trp Thr	Gly Phe Asp Phe Ser
130	135	140
Gly Arg Asn Asn Ala Tyr	Ser Asp Phe Lys Trp Arg Trp Phe His Phe	
145	150	155 160
Asn Gly Val Asp Trp Asp Gln Arg Tyr	Gln Glu Asn His Ile Phe Arg	
165	170	175
Phe Ala Asn Thr Asn Trp Asn Trp Arg Val	Asp Glu Glu Asn Gly Asn	
180	185	190
Tyr Asp Tyr Leu Leu Gly Ser	Asn Ile Asp Phe Ser His Pro Glu Val	
195	200	205
Gln Asp Glu Leu Lys Asp Trp Gly Ser Trp Phe Thr	Asp Glu Leu Asp	
210	215	220
Leu Asp Gly Tyr Arg Leu Asp Ala Ile Lys His Ile Pro Phe Trp Tyr		
225	230	235 240
Thr Ser Asp Trp Val Arg His Gln Arg	Asn Glu Ala Asp Gln Asp Leu	
245	250	255
Phe Val Val Gly Glu Tyr Trp Lys Asp Asp Val Gly Ala Leu Glu Phe		
260	265	270
Tyr Leu Asp Glu Met Asn Trp Glu Met Ser Leu Phe Asp Val Pro Leu		
275	280	285
Asn Tyr Asn Phe Tyr Arg Ala Ser Gln Gln Gly Gly Ser Tyr Asp Met		
290	295	300
Arg Asn Ile Leu Arg Gly Ser Leu Val Glu Ala His Pro Met His Ala		
305	310	315 320

Val Thr Phe Val Asp Asn His Asp Thr Gln Pro Gly Glu Ser Leu Glu
325 330 335

Ser Trp Val Ala Asp Trp Phe Lys Pro Leu Ala Tyr Ala Thr Ile Leu
340 345 350

Thr Arg Glu Gly Gly Tyr Pro Asn Val Phe Tyr Gly Asp Tyr Tyr Gly
355 360 365

Ile Pro Asn Asp Asn Ile Ser Ala Lys Lys Asp Met Ile Asp Glu Leu
370 375 380

Leu Asp Ala Arg Gln Asn Tyr Ala Tyr Gly Thr Gln His Asp Tyr Phe
385 390 395 400

Asp His Trp Asp Val Val Gly Trp Thr Arg Glu Gly Ser Ser Ser Arg
405 410 415

Pro Asn Ser Gly Leu Ala Thr Ile Met Ser Asn Gly Pro Gly Gly Ser
420 425 430

Lys Trp Met Tyr Val Gly Arg Gln Asn Ala Gly Gln Thr Trp Thr Asp
435 440 445

Leu Thr Gly Asn Asn Gly Ala Ser Val Thr Ile Asn Gly Asp Gly Trp
450 455 460

Gly Glu Phe Phe Thr Asn Gly Gly Ser Val Ser Val Tyr Val Asn Gln
465 470 475 480

<210> 20

<211> 1441

<212> DNA

<213> *Bacillus clausii* KSM-K16

<220>

<221> CDS

<222> (164)..(1303)

<223>

<400> 20

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acttaatggt aataattggt tcccaatagg caaatctttc taactttgat acgtttaaac	120
taccagcttg gacgagttgg gataaaagtg aggagggaac cga atg aag aaa ccg	175
Met Lys Lys Pro	
1	
ttg ggg aaa att gtc gca agc acc gca cta ctc att tct gtt gct ttt	223
Leu Gly Lys Ile Val Ala Ser Thr Ala Leu Leu Ile Ser Val Ala Phe	
5 10 15 20	
agt tca tcg atc gca tcg gct gct gag gaa gca aaa gaa aaa tat tta	271
Ser Ser Ser Ile Ala Ser Ala Ala Glu Glu Ala Lys Glu Lys Tyr Leu	
25 30 35	
att ggc ttt aat gag cag gaa gca gtt agt gag ttt gta gag caa ata	319
Ile Gly Phe Asn Glu Gln Glu Ala Val Ser Glu Phe Val Glu Gln Ile	
40 45 50	
gag gca aat gac gat gtc gcg att ctc tct gag gaa gag gaa gtc gaa	367
Glu Ala Asn Asp Asp Val Ala Ile Leu Ser Glu Glu Glu Glu Val Glu	
55 60 65	
att gaa ttg ctt cat gag ttt gaa acg att cct gtt tta tct gtt gag	415
Ile Glu Leu Leu His Glu Phe Glu Thr Ile Pro Val Leu Ser Val Glu	
70 75 80	
tta agt cca gaa gat gtg gac gcg ctt gag ctc gat cca acg att tcg	463
Leu Ser Pro Glu Asp Val Asp Ala Leu Glu Leu Asp Pro Thr Ile Ser	
85 90 95 100	
tat att gaa gag gat gca gaa gta acg aca atg gcg caa tca gtg cca	511
Tyr Ile Glu Glu Asp Ala Glu Val Thr Thr Met Ala Gln Ser Val Pro	
105 110 115	
tgg gga att agc cgt gta caa gcc cca gct gcc cat aac cgt gga ttg	559
Trp Gly Ile Ser Arg Val Gln Ala Pro Ala Ala His Asn Arg Gly Leu	
120 125 130	
aca ggt tct ggt gta aaa gtt gct gtc ctc gat acg ggt att tcc acc	607
Thr Gly Ser Gly Val Lys Val Ala Val Leu Asp Thr Gly Ile Ser Thr	
135 140 145	
cat cca gac tta aat att cgc ggt ggt gct agc ttt gtg cca gga gaa	655

His Pro Asp Leu Asn Ile Arg Gly Gly Ala Ser Phe Val Pro Gly Glu	
150 155 160	
cca tcc act caa gat gga aat gga cat ggc acg cat gtg gca ggg acg	703
Pro Ser Thr Gln Asp Gly Asn Gly His Gly Thr His Val Ala Gly Thr	
165 170 175 180	
att gct gct tta aac aat tcg att ggc gtt ctg ggc gta gca ccg agc	751
Ile Ala Ala Leu Asn Asn Ser Ile Gly Val Leu Gly Val Ala Pro Ser	
185 190 195	
gcg gaa cta tac gct gta aaa gta tta ggc gcg agc ggt tca ggt tcg	799
Ala Glu Leu Tyr Ala Val Lys Val Leu Gly Ala Ser Gly Ser Gly Ser	
200 205 210	
gtc agc tcg att gcc caa gga ttg gaa tgg gca ggg aac aat ggc atg	847
Val Ser Ser Ile Ala Gln Gly Leu Glu Trp Ala Gly Asn Asn Gly Met	
215 220 225	
cac gtt gcg aat ttg agt tta gga agc ccg tcg ccg agt gca aca ctt	895
His Val Ala Asn Leu Ser Leu Gly Ser Pro Ser Pro Ser Ala Thr Leu	
230 235 240	
gag caa gct gtt aat agc gct act tct aga ggc gtt ctt gtc gta gca	943
Glu Gln Ala Val Asn Ser Ala Thr Ser Arg Gly Val Leu Val Val Ala	
245 250 255 260	
gca tct ggt aat tca ggt gca ggc tca atc agc tat ccg gcc cgt tat	991
Ala Ser Gly Asn Ser Gly Ala Gly Ser Ile Ser Tyr Pro Ala Arg Tyr	
265 270 275	
gcg aac gca atg gca gtc gga gcg act gac caa aac aac aac cgc gct	1039
Ala Asn Ala Met Ala Val Gly Ala Thr Asp Gln Asn Asn Asn Arg Ala	
280 285 290	
agc ttt tca cag tat gga gct ggg ctt gac att gtc gcg cca ggt gtc	1087
Ser Phe Ser Gln Tyr Gly Ala Gly Leu Asp Ile Val Ala Pro Gly Val	
295 300 305	
aat gtg cag agc aca tac cca ggt tca aca tat gcc agc tta aac ggt	1135
Asn Val Gln Ser Thr Tyr Pro Gly Ser Thr Tyr Ala Ser Leu Asn Gly	
310 315 320	
aca tcg atg gct act cct cat gtt gca ggt gta gca gcc ctt gtt aaa	1183
Thr Ser Met Ala Thr Pro His Val Ala Gly Val Ala Ala Leu Val Lys	
325 330 335 340	
caa aag aat cca tct tgg tcc aat gta caa atc cgc aat cat cta aag	1231
Gln Lys Asn Pro Ser Trp Ser Asn Val Gln Ile Arg Asn His Leu Lys	

345	350	355	
aat acg gca acg ggt tta gga aac acg aac ttg tat gga agc ggg ctt			1279
Asn Thr Ala Thr Gly Leu Gly Asn Thr Asn Leu Tyr Gly Ser Gly Leu			
360	365	370	
gtc aat gca gaa gcg gca aca cgc taatcaataa taataacgct gtgtgcttta			1333
Val Asn Ala Glu Ala Ala Thr Arg			
375	380		
agcacacagc gttttttagt gtgtatgaat cgaaaaagag aaatagatcg ctgatttcaa			1393
aaagcgagcg taaagggcta ttgaagctct ttacgcttgc aggatttg			1441

<210> 21
 <211> 380
 <212> PRT
 <213> *Bacillus clausii* KSM-K16

<400> 21

Met	Lys	Lys	Pro	Leu	Gly	Lys	Ile	Val	Ala	Ser	Thr	Ala	Leu	Leu	Ile
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Ser	Val	Ala	Phe	Ser	Ser	Ser	Ile	Ala	Ser	Ala	Ala	Glu	Glu	Ala	Lys
		20						25					30		

Glu	Lys	Tyr	Leu	Ile	Gly	Phe	Asn	Glu	Gln	Glu	Ala	Val	Ser	Glu	Phe
		35					40					45			

Val	Glu	Gln	Ile	Glu	Ala	Asn	Asp	Asp	Val	Ala	Ile	Leu	Ser	Glu	Glu
	50					55					60				

Glu	Glu	Val	Glu	Ile	Glu	Leu	Leu	His	Glu	Phe	Glu	Thr	Ile	Pro	Val
65					70					75				80	

Leu	Ser	Val	Glu	Leu	Ser	Pro	Glu	Asp	Val	Asp	Ala	Leu	Glu	Leu	Asp
				85					90					95	

Pro	Thr	Ile	Ser	Tyr	Ile	Glu	Glu	Asp	Ala	Glu	Val	Thr	Thr	Met	Ala
					100			105					110		

Gln Ser Val Pro Trp Gly Ile Ser Arg Val Gln Ala Pro Ala Ala His
115 120 125

Asn Arg Gly Leu Thr Gly Ser Gly Val Lys Val Ala Val Leu Asp Thr
130 135 140

Gly Ile Ser Thr His Pro Asp Leu Asn Ile Arg Gly Gly Ala Ser Phe
145 150 155 160

Val Pro Gly Glu Pro Ser Thr Gln Asp Gly Asn Gly His Gly Thr His
165 170 175

Val Ala Gly Thr Ile Ala Ala Leu Asn Asn Ser Ile Gly Val Leu Gly
180 185 190

Val Ala Pro Ser Ala Glu Leu Tyr Ala Val Lys Val Leu Gly Ala Ser
195 200 205

Gly Ser Gly Ser Val Ser Ser Ile Ala Gln Gly Leu Glu Trp Ala Gly
210 215 220

Asn Asn Gly Met His Val Ala Asn Leu Ser Leu Gly Ser Pro Ser Pro
225 230 235 240

Ser Ala Thr Leu Glu Gln Ala Val Asn Ser Ala Thr Ser Arg Gly Val
245 250 255

Leu Val Val Ala Ala Ser Gly Asn Ser Gly Ala Gly Ser Ile Ser Tyr
260 265 270

Pro Ala Arg Tyr Ala Asn Ala Met Ala Val Gly Ala Thr Asp Gln Asn
275 280 285

Asn Asn Arg Ala Ser Phe Ser Gln Tyr Gly Ala Gly Leu Asp Ile Val
290 295 300

Ala Pro Gly Val Asn Val Gln Ser Thr Tyr Pro Gly Ser Thr Tyr Ala
 305 310 315 320

Ser Leu Asn Gly Thr Ser Met Ala Thr Pro His Val Ala Gly Val Ala
 325 330 335

Ala Leu Val Lys Gln Lys Asn Pro Ser Trp Ser Asn Val Gln Ile Arg
 340 345 350

Asn His Leu Lys Asn Thr Ala Thr Gly Leu Gly Asn Thr Asn Leu Tyr
 355 360 365

Gly Ser Gly Leu Val Asn Ala Glu Ala Ala Thr Arg
 370 375 380

- <210> 22
- <211> 46
- <212> DNA
- <213> Artificial

<220>

<223> Oligonucleotide as PCR primer; its 3'-portion designed from nucleotide sequence of the alkaline protease gene in *Bacillus clausii* KSM-K16 and its 5'-portion designed from nucleotide sequence of the upstream region of the alkaline cellulase gene in *Bacillus* sp. KSM-S237

<400> 22
 actttaaaaa tatttaggag gtaatatgaa gaaaccgttg gggaaa 46

- <210> 23
- <211> 32
- <212> DNA
- <213> Artificial

<220>

<223> Oligonucleotide as PCR primer designed from nucleotide sequence of the downstream region of the alkaline protease gene in *Bacillus clausii* KSM-K16 with a insertion of the BglII restriction site at the 5'-end

<400> 23
 ggagatctt cagcgatcta tttctctttt tc 32

<210> 24
<211> 25
<212> DNA
<213> Artificial

<220>

<223> Oligonucleotide as PCR primer designed from nucleotide sequence of the upstream region of the alkaline cellulase gene in *Bacillus* sp. KSM-S237 with a insertion of the BamHI restriction site at the 5'-end

<400> 24
cccggatcca acaggcttat attta

25

<210> 25
<211> 46
<212> DNA
<213> Artificial

<220>

<223> Oligonucleotide as PCR primer; its 3'-portion designed from nucleotide sequence of the upstream region of the alkaline cellulase gene in *Bacillus* sp. KSM-S237 and its 5'-portion designed from nucleotide sequence of the alkaline protease gene in *Bacillus clausii* KSM-K16

<400> 25
tttccccaac gggtttcttca tattacctcc taaatatttt taaagt

46

<210> 26
<211> 30
<212> DNA
<213> Artificial

<220>

<223> Oligonucleotide as PCR primer; its 3'-portion designed from nucleotide sequence of the alkaline amylase gene in *Bacillus* sp. KSM-K38 and its 5'-portion designed from nucleotide sequence of the alkaline cellulase gene in *Bacillus* sp. KSM-S237

<400> 26
gctcttgcag cagatggatt gaacggtacg

30

<210> 27
<211> 30
<212> DNA
<213> Artificial

<220>

<223> Oligonucleotide as PCR primer designed from nucleotide sequence of the downstream region of the alkaline amylase gene in Bacillus sp. KSM-K38 with a insertion of the XbaI restriction site at the 5'-end

<400> 27

ttggtctaga cccaagctt caaagtcgta

30

<210> 28
<211> 29
<212> DNA
<213> Artificial

<220>

<223> Oligonucleotide as PCR primer: its 3'-portion designed from nucleotide sequence of the alkaline cellulase gene in Bacillus sp. KSM-S237 and its 5'-portion designed from nucleotide sequence of the alkaline amylase gene in Bacillus sp. KSM-K38

<400> 28

ttcaatccat ctgctgcaag agctgccg

29